

ABSTRACT

Enterococci are the fast emerging organisms causing serious nosocomial outbreaks due to the virulence factors and multiple drug resistance pattern acquired by the enterococcal species. They are intrinsically resistant to antibiotics like cephalosporins, clindamicin, co-trimoxazole and low level resistant to aminoglycosides. Optimal antimicrobial treatment for serious enterococcal infections requires combinations of a cell wall-active agent, such as a penicillin, Ampicillin or a glycopeptide, with an aminoglycoside results in synergistic action which results in bactericidal activity against this organism. The recommended aminoglycosides for treating serious enterococcal infections are Gentamicin and Streptomycin. The addition of an agent that interferes with cell wall synthesis, such as ampicillin (or vancomycin), markedly increases uptake of the aminoglycoside, greatly enhance the killing of the Enterococcus. But when Enterococci have acquired high resistance to aminoglycosides due to the presence of resistant determining genes that mediate production of aminoglycoside modifying enzymes which eliminates synergistic bactericidal effect.

Aims and objectives:

This study was carried out to isolate and speciate Enterococcus from various samples collected from Govt rajaji hospital, Madurai, to find out prevalence of High level gentamicin resistance among the isolated enterococcal species by phenotypic screening methods. (Disc diffusion method (gentamicin 120 ug disc), E-test and agar dilution) and detecting the genes responsible for high level gentamicin resistance by molecular technique (polymerase chain reaction) among isolated

Materials and Methods: High level gentamicin resistance among isolated enterococci were screened by disc diffusion method, agar dilution method and E-

test method and detection of genes responsible for high level gentamicin resistance by polymerase chain reaction.

Results: *E. faecalis* was the predominant species isolated in the present study with an isolation rate of about 77/104(74.03%), followed by *E. faecium* 27/104(25.96%). Other species of enterococci were not isolated. High level gentamicin resistance were seen in 63.4% (66 out of 104) of Enterococcal isolates by disc diffusion method and similar results were also seen in E-test method & agar dilution method. All the 66 high level gentamicin resistance isolates (*E. faecalis*-48 and *E. faecium*-18) were subjected to Polymerase Chain Reaction for the detection of resistance determining genes *aac(6)-Ie-aph(2)-Ia*, *aph(2)-Ib*, *aph(2)-Ic*, *aph(2)-Id*. In PCR assay, out of four genes, the gene *aac(6)-Ie-aph(2)-Ia* was alone detected in all the isolates. This gene codes the bifunctional aminoglycoside modifying enzyme which confers resistance to all the commonly used aminoglycosides except streptomycin. The present study showed the *aac(6)-Ie-aph(2)-Ia gene* was the most prevalent gene present in Enterococci.

Conclusion: The higher prevalence of High level gentamicin resistant enterococci have posed a serious problem in the management of serious enterococcal infection. This type of serious infections is usually treated with aminoglycosides (gentamicin or streptomycin) along with the cell wall acting agents, thus limiting the therapeutic options. Judicious use of aminoglycosides, and regular surveillance of all Enterococcal isolates for High level gentamicin resistance is necessary for the prevention of nosocomial transmission of resistant strains. Appropriate surveillance, stringent infection control practice and hospital infection control committee guidance is very important to control the spread of High level gentamicin resistant Enterococci.

Keywords: high level gentamicin resistance, polymerase chain reaction, antibiotic susceptibility testing,

KEYWORDS

HIGH LEVEL GENTAMICIN RESISTANCE

HIGH LEVEL STREPTOMYCIN RESISTANCE

ANTIBIOTIC SUSCEPTIBILITY TESTING

VANCOMYCIN RESISTANT ENREROCOCCI

VANCOMYCIN SENSITIVE ENREROCOCCI

MINIMUM INHIBITORY CONCENTRATION

BRAIN HEART INFUSION AGAR

MUELLER HINTON AGAR

BLOOD AGAR PLATE

NUTRIENT AGAR

BILE ESCULIN AGAR

POLYMERASE CHAIN REACTION